

# Enzyme-linked immunosorbent assay (ELISA)

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 An abbreviated version of this protocol was published in Science in Aug 2022

GPNMB confers risk for Parkinson's disease through interaction with  $\alpha$ -synuclein

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## Detailed protocol

### Enzyme-Linked Immunosorbent Assay (ELISA):

GPNMB protein levels within human plasma and CSF samples were measured with ELISA kits (R&D systems, [https://www.rndsystems.com/products/human-osteoactivin-gpnmb-duoset-elisa\\_dy2550](https://www.rndsystems.com/products/human-osteoactivin-gpnmb-duoset-elisa_dy2550)) according to manufacturer's instructions (we did not modify from instructions). CSF and plasma samples were diluted by factors of 1 in 2 (CSF) and 1 in 30 (plasma) to obtain optical density measurements within the standard range. All samples were run in duplicates and absorbance at 450nm was determined by a microplate reader (Berthold Technologies, Tristar LB 941). Only duplicate samples with a coefficient of variation (CV) <25% were retained for analysis, and the average CV across all samples used was 3.3%. Moreover, replicate samples assayed by ELISA on different days, by different operators, across multiple freeze-thaw cycles, demonstrated excellent reproducibility (Pearson  $r=0.97$ ).

All samples are from our central biobank, whose protocols can be found here: <https://sites.google.com/site/chenplotkinlab/biobank>.

**How to cite:** (Readers should cite both the Bio-protocol preprint and the original research article where this protocol was used)

1. Diaz-Ortiz, M. and Chen-Plotkin, A. (2022). Enzyme-linked immunosorbent assay (ELISA). Bio-protocol Preprint. [bio-protocol.org/prep1888](https://bio-protocol.org/prep1888).
2. Diaz-Ortiz, M. E., Seo, Y., Posavi, M., Carceles Cordon, M., Clark, E., Jain, N., Charan, R., Gallagher, M. D., Unger, T. L., Amari, N., Skrinak, R. T., Davila-Rivera, R., Brody, E. M., Han, N., Zack, R., Van Deerlin, V. M., Tropea, T. F., Luk, K. C., Lee, E. B., Weintraub, D. and Chen-Plotkin, A. S. (2022). GPNMB confers risk for Parkinson's disease through interaction with  $\alpha$ -synuclein. Science 377(6608). DOI: [10.1126/science.abk0637](https://doi.org/10.1126/science.abk0637)

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